ELSEVIER

The Science of the Total Environment 284 (2002) 237–247

the Science of the Total Environment
An International Journal for Scientific Research into the Environment and its Relationship with Man

www.elsevier.com/locate/scitotenv

Partition coefficients for the trihalomethanes among blood, urine, water, milk and air

Stuart Batterman*, Lian Zhang, Shugin Wang, Alfred Franzblau

Department of Environmental Health Sciences, University of Michigan, 109 Observatory Drive, Ann Arbor, MI 48109-2029, USA

Received 13 December 2000; accepted 7 May 2001

Abstract

Chloroform, bromodichloromethane, chlorodibromomethane, and bromoform comprise the trihalomethanes, a group of widespread and mildly lipophilic compounds that result from water chlorination and other sources. Many animal studies show the chronic toxicity and carcinogenicity of these compounds, and recent work has demonstrated the importance of both ingestion and inhalation exposure pathways. This study presents partition coefficients describing the equilibrium among biological compartments (air, water, blood, milk, urine) for the four THMs based on results of headspace–gas chromatographic analyses performed under equilibrium conditions and at 37°C. The calculated partition coefficients ranged from 2.92 to 4.14 for blood/water, 1.54–2.85 for milk/blood, and 3.41–4.93 for blood/urine, with the lowest being chloroform and the highest being bromoform. Both human and cow milk were tested, with similar results. The available samples of human milk may not fully account for differences in lipid content and possibly other factors that affect estimates of partition coefficients. Simultaneous measurements of milk and blood in exposed individuals are suggested to confirm laboratory results. Partition coefficients are predicted using the octanol–air partition coefficient, also measured in this study, and the octanol–water partition coefficient. Results are similar to literature estimates for liquid/air partitioning of chloroform and chlorodibromomethane, but they differ from predictions based on hydrophobicity and lipid content. High correlations between the derived partitioned coefficients and the molecular structure (number of Br atoms)(and physical properties molecular weight and boiling point) are found for these analogous chemicals. In humans, THMs are both stored and metabolized with relatively rapid clearance rates. The derived partition coefficients can help to interpret results of biological monitoring and predict the potential for the accumulation and transfer of chemicals, specifically by the application of physiologically-based pharmacokinetic models. THM exposures to potentially susceptible populations, e.g. nursing infants, can be predicted using either such models. © 2002 Elsevier Science B.V. All rights reserved.

Keywords: Biological monitoring; Blood; Chloroform; Disinfection by-products; Exposure; Milk; Partitioning; Trihalomethanes

* Corresponding author. Tel.: +1-734-763-2417; fax: +1-734-764-9424.
E-mail address: stuartb@umich.edu (S. Batterman).

0048-9697/02/$ - see front matter © 2002 Elsevier Science B.V. All rights reserved.
PII: S0048-9697(01)00890-7

1. Introduction

Chlorination has been used for water disinfection since the last century, and it remains the most widely used technology in the water industry. The finding that trihalomethanes (THMs), namely chloroform ($CHCl_3$), bromodichloromethane ($CHCl_2Br$), chlorodibromomethane ($CHClBr_2$) and bromoform ($CHBr_3$), are formed during water chlorination has led to extensive research on the occurrence of these chemicals and possible adverse health effects. In chlorinated drinking water, the total THM concentration (sum of $CHCl_3$, $CHCl_2Br$, $CHClBr_2$ and $CHBr_3$) can range from tens up to hundreds of μg/l (McGuire and Meadow, 1988; Ayotte et al., 1989; McGuire, 1989; Craun, 1990; Cumming and Jolley, 1993; Arora et al., 1997). Toxicologic studies of these disinfection byproducts (DBPs) using mammalian animal models have shown chronic toxicity and carcinogenicity, for example, kidney cancers in rats are associated with $CHCl_3$ (NCI, 1976; Jorgenson et al., 1985); liver tumors in female $B6C3F_1$ mice are associated with $CHClBr_2$ (NTP, 1985); tumors in various animals are associated with $CHCl_2Br$ (NTP, 1986, 1987); and clear evidence of carcinogenicity of $CHBr_3$ is found in female rats and some evidence in the male (NTP, 1988). In consequence, US EPA (1997a) has assigned, in a weight-of-evidence classification, $CHCl_3$, $CHCl_2Br$ and $CHBr_3$ as probable human carcinogens, and $CHClBr_2$ as a possible human carcinogen. The US EPA currently imposes an MCL of 80 μg/l for total THMs in drinking water, and limits of 40 μg/l have been proposed. Some European countries have restricted total THMs to as low as 10 μg/l (Germany).

For adults, exposure pathways that can lead to potentially significant uptake of THMs include the ingestion of tap water and contaminated foods or drinks, inhalation of vapor phase THMs resulting from showering, dish washing and swimming, and dermal exposure during bathing, showering, washing and swimming (Filser and Bolt, 1981; Jo et al., 1990; Maxwell et al., 1991; McKone, 1993; Olin, 1998). For infants, exposure pathways may differ due to the potential significance of lactational transfer from mother to infant. The exposure route may affect the rate of metabolism and the toxicity of the compound (Maxwell et al., 1991; Weisel and Jo, 1996).

Halogenated organic chemicals like the THMs tend to be fat soluble and hydrophobic, thus they accumulate into fatty substances, e.g. the lipid fraction of blood or milk. Measurements of human milk over nearly 50 years indicate the magnification of a diverse set of compounds, including ketones, aromatics, aldehydes, halogenated compounds, trace metals and drugs. Pellizzari et al. (1982) reported qualitative identification of many compounds in breast milk, including $CHCl_3$ and $CHClBr_2$. The importance of lactating mothers' exposure to lipophilic and persistent chemicals like polychlorinated biphenyls (PCBs), pesticides and volatile organic compounds is well known, both in terms of chemical exposure (body burden) (Newsome et al., 1995; Dewailly et al., 1996; Craan and Haines, 1998) and health impacts on the infant (Gladen et al., 1988; Koppe et al., 1989; Jacobson et al., 1990; Byckowski et al., 1994). Other contaminants in human milk have been measured or estimated to reach levels of concern due to maternal inhalation exposures in the home (Schrieber, 1993) and workplace e.g. Byckowski et al., 1994; Fisher et al., 1997), and from maternal ingestion of persistent environmental contaminants (e.g. Sim and McNeil, 1992). Human milk can provide an excellent biological indicator of infant exposure to lipophilic substances (Sim and McNeil, 1992; Craan and Haines, 1998). For the THMs, the information available regarding partitioning in biological media is incomplete, yet critically important in understanding biological monitoring results and predicting exposures.

This paper presents experimentally determined partition coefficients for the four THMs among human blood, milk, urine, saline and air at environmentally-relevant concentrations. Results for human and cow milk are compared. Experimental results are compared to the literature, including several partitioning models.

2. Background

2.1. Experimental determination of partition coefficients

Partition coefficients are most simply determined using the vial equilibrium method in which a known quantity of a chemical is injected as a liquid or vapor into a vial partially filled with the fluid of interest. After equilibration at the temperature of interest (typically 37°C), the chemical in vapor phase in the headspace is quantified, establishing the liquid/air partition coefficient for that fluid. Partition coefficients for two fluids, say milk/blood, may be derived as the ratio of independent determinations of the liquid/air partition coefficients for each fluid. Using the vial equilibrium method and vapor injections, Fisher et al. (1997) obtained human blood/air, milk/air, and milk/blood partition coefficients for $CHCl_3$ of 10.68, 16.24 and 1.5, respectively, based on nine samples and very high concentrations (initially 1000 ppm in headspace). Gargas et al. (1989) obtained rat blood/air partition coefficients of 20.8 for $CHCl_3$ and 116 for $CHClBr_2$, and human blood/air partition coefficients of 6.85 for $CHCl_3$ and 52.7 for $CHClBr_2$, respectively. The concentrations used in these tests, which approach solubility limits, may be relevant to very high exposures, possibly those encountered in unusual occupational settings. Partition coefficients at environmentally relevant concentrations have not been published. Partition coefficients for the two other THMs ($CHCl_2Br$ and $CHBr_3$), which are expected to be higher (see below), also have not been published.

While simple, there are subtleties to the vial headspace method. For example, Gargas et al. (1989) compared vapor injections into the headspace with liquid injections into the liquid phase [as used by Sato and Nakajima (1979)]. The latter method tended to inflate liquid/air partition coefficients by 20–50%, indicating that the chemicals in the liquid and gaseous phases in the vial did not reach equilibrium.

2.2. Predicting partition coefficients

There are several ways to predict partition coefficients. Using the octanol–air partition coefficient ($\log K_{OA}$), which describes the distribution of chemicals between air and environmental lipids (Harner and Bidleman, 1996), and the lipid content (f_L) (of the liquid of concern blood, milk, etc.), the partition coefficient for liquid/air ($K_{L/A}$) may be estimated as:

$$K_{L/A} = f_L K_{OA} \quad (1)$$

[Partition coefficients here are expressed on a mass/volume basis, that is $K_{L/A} = (mg/l)_{liquid}/(mg/l)_{air}$.] In this equation, it is assumed that the chemical partitions only into the lipid fraction, and that different lipids have essentially the same partitioning characteristics as octanol. (As shown later, these assumptions appear problematic for blood and other fluids.) K_{OA} can be either experimentally determined or calculated as the product of the octanol/water partition coefficient (K_{OW}) and the water/air partition coefficient ($K_{W/A}$), thus, $\log K_{OA} = \log K_{OW} + \log K_{W/A}$. The latter approach tends to underestimate K_{OA} (Harner and Bidleman, 1996).

The more traditional approach for estimating the partition coefficient amongst two liquids uses the lipid fractions of the two liquids:

$$K_{X/Y} = [(1 - f_{LX}) + f_{LX} K_{OW}] / [(1 - f_{LY}) + f_{LY} K_{OW}] \quad (2)$$

where $K_{X/Y}$ is the partition coefficient for liquids x and y and f_{LX} and f_{LY} are the lipid contents of fluids x and y, respectively (Gargas et al., 1989). Again, this approach assumes that the lipophilicity of the liquid is comparable to that of octanol. Additionally, the non-lipid fraction is assumed to be water, and all concentrations are assumed to be below solubility limits.

Most of the parameters required in Eqs. (1) and (2) are available. The total lipids content in blood is from 0.5 to 0.7% (Albritton, 1952; Van-

der et al., 1990; Guyton, 1991). (Blood lipids include cholesterol esters, free cholesterol, phospholipid, and neutral fat.) In human milk, estimates of the mean lipid fraction range from 3.3 to 5.0% (Ryan et al., 1991; Guyton, 1991; US EPA, 1997b). Published values of log K_{OW} are 1.97 for $CHCl_3$, 2.10 for $CHCl_2Br$, 2.24 for $CHClBr_2$ and 2.38 for $CHBr_3$ (Mabe et al., 1982; Hansch and Leo, 1985). Because the K_{OA} method is quite recent, relatively few determinations of K_{OA} are available in the literature, and none was found for the THMs.

3. Materials and methods

3.1. Materials

Whole blood was obtained from the American Red Cross (Southeastern Michigan Region Branch). The sample was anticoagulated by adding citrate phosphate dextrose adenine solution, USP, and stored at 4°C.

Human milk was obtained from Wake Med Lactation Center and Milk Bank in Raleigh, NC. The raw milk was donated by five mothers. The milk was stored frozen.

Whole cow milk was purchased from two major local supermarkets.

Urine was collected from volunteers and pooled.

Saline was prepared by dissolving 9 g of NaCl in 1 l of distilled water.

THM standards included a mixture of $CHCl_3$, $CHCl_2Br$, $CHClBr_2$ and $CHBr_3$ prepared at 2000 μg/l each in methanol (Trihalomethanes Calibration Mix, Supelco) and neat standards of the same compounds (Supelco, except chloroform from Fluka). Stock solutions at 200 μg/ml were prepared in methanol and further diluted to prepare calibration and additive solutions.

Anhydrous octanol (99 + %) was purchased from Aldrich.

3.2. Experimental methods

Partition coefficients were determined for the four THMs in human blood, human milk, cow milk, human urine, saline and distilled water using headspace–gas chromatographic analysis, following Gargas et al. (1989) except that the THMs (in methanol solution) were injected as 1–2-μl liquids on the walls of the septum-sealed vials containing 5 ml of the sample solution. Methanol introduced in the injection accounted for only 0.02–0.04% of the fluid in the vial, and thus should not affect the accuracy of the determination (see Section 4). A portion of the THM solution evaporated upon introduction; the remainder was then swirled into the liquid phase. Experiments were performed to evaluate the time needed to reach equilibrium, and to measure pre-existing (background) levels of the THMs in the blood, urine, human and cow milk samples.

THM concentrations were measured using a Tekmar 7000 headspace autosampler, which provided both phase equilibration and gas phase sampling, coupled to a Varian 3700 gas chromatograph equipped with a linearized electron capture detector, 12-bit data acquisition system, and Chrome (Laboratory Technologies Corporation) software for quantification. Known quantities of THMs were injected into 22-ml sealed vials containing known volumes of liquids of interest. To equilibrate, the vials were incubated at 37°C in the autosampler for 1.5 h, a time sufficient to reach equilibrium (see below). The vial was shaken at low power (without wetting the septum at power setting 3) for 2 min prior to sampling. Using a heated sample loop, 1 ml of vial headspace was injected into the GC column (2.0 m × 4 mm i.d. stainless steel column packed with 20% THEED on 60/90 mesh firebrick). The oven temperature was held at 40°C for 2 min, ramped at 20°C/min to 75°C, and held for 1 min. To improve quantification of $CHCl_3$, a separate program, a constant oven temperature of 30°C, was used. The method detection limits for $CHCl_3$, $CHCl_2Br$, $CHClBr_2$ and $CHBr_3$ were 0.1, 0.03, 0.04 and 0.5 μg/l, respectively. All determinations were carried out within the linear range of the instrument. The system was calibrated using gas standards prepared in empty autosampler vials.

Concentrations of THMs in the liquid phase, C_L (ng/ml), were calculated as:

$$C_L = (Q - C_A[22 - V_L])/V_L \quad (3)$$

where Q = amount of THMs injected into the vial (ng); C_A = measured gas phase concentration of THMs (ng/ml); and V_L = volume of liquid phase in the 22-ml vial (ml). The liquid/air partition coefficient was calculated as:

$$K_{L/A} = C_L/C_A \quad (4)$$

Eqs. (3) and (4) are equivalent to that used by Gargas et al. (1989) to calculate liquid/air partition coefficients. Partition coefficients for fluids x and y (e.g. milk/blood) were derived as:

$$K_{X/Y} = (C_{LX}/C_{AX})/(C_{LY}/C_{AY}) = K_{X/A}/K_{Y/A} \quad (5)$$

K_{OA} (octanol/air) values were determined using the same vial equilibrium method except that a small volume (100–200 μl) of octanol was used so that sufficient fractions of THMs remained in the headspace for quantification.

4. Results

4.1. Equilibration time and background levels

The vials approached equilibrium conditions within 90 min for all liquids and chemicals tested (Fig. 1). After this period, for example, THM concentrations in the headspace over milk reached ≥ 95% of those achieved after 180 min. Similar results were obtained for other liquids tested. The following reports results using an equilibration time of 90 min and a temperature of 37°C.

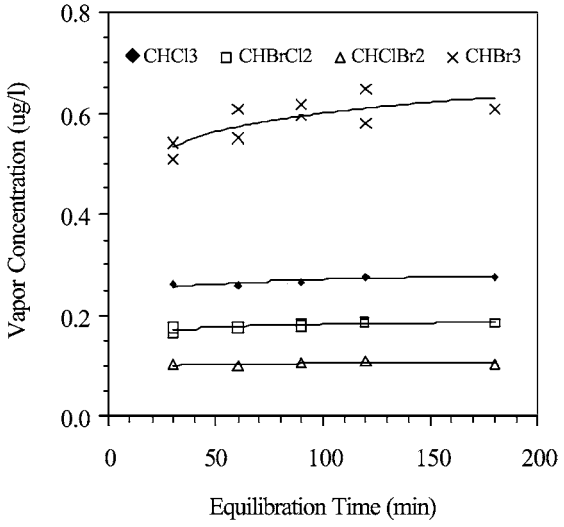

Fig. 1. Effect of sample equilibration time on headspace concentrations of THMs over human milk.

Using headspace tests, no THMs were detected in samples of blood, urine and human milk before the partition experiments. However, the store-bought cow milk had $CHCl_3$ concentrations that ranged from 1 to 3 μg/l, depending on the sample. No other THMs in the cow milk samples were detected. The $CHCl_3$ background was taken into account in determining the $CHCl_3$ partition coefficient.

4.2. Partition coefficients

Partition coefficients experimentally determined for blood/air, milk/air, urine/air, water/air and saline/air are listed in Table 1, while

Table 1
Liquid–air partition coefficients for human milk, blood, urine and saline

THM	$K_{B/A}$ blood/air	$K_{M/A}$ milk/air	$K_{U/A}$ urine/air	$K_{S/A}$ saline/air	$K_{W/A}$ water/air
$CHCl_3$	10.7 (0.7)	16.5 (5.0)	3.14 (0.36)	3.60 (0.05)	3.66 (0.31)
$CHCl_2Br$	26.6 (1.4)	33.5 (5.0)	6.44 (0.56)	6.23 (0.10)	7.43 (0.35)
$CHClBr_2$	49.2 (2.3)	89.3 (13.4)	11.20 (1.11)	11.26 (0.07)	11.83 (0.71)
$CHBr_3$	102.3 (8.8)	291.5 (42.2)	20.74 (3.34)	24.95 (1.66)	24.71 (4.00)

S.D. in parentheses. Sample size $n = 10$ for $K_{B/A}$; $n = 16$ for $K_{M/A}$, except $CHCl_3$ where $n = 34$; $n = 5$ for $K_{U/A}$; and $n = 4$ for $K_{W/A}$. All coefficients here and in subsequent tables expressed as $(mg/l)_{liquid}/(mg/l)_{air}$.

Table 2
Liquid–liquid partition coefficients based on experimental measurements and Eq. (5)

THM	$K_{B/W}$ blood/water	$K_{M/B}$ milk/blood	$K_{B/U}$ blood/urine
$CHCl_3$	2.92	1.54	3.41
$CHCl_2Br$	3.58	1.26	4.13
$CHClBr_2$	4.16	1.82	4.39
$CHBr_3$	4.14	2.85	4.93

partition coefficients calculated using Eq. (5) for blood/water, milk/blood, and blood/urine are listed in Table 2. The blood/air, milk/air and milk/blood partition coefficients for $CHCl_3$ (10.72, 16.53 and 1.54, respectively) are nearly identical to those reported by Fisher et al. (1997), namely, 10.68, 16.24 and 1.52, respectively. The measured blood/air partition coefficient for $CHClBr_2$ (49.2) was also close to that (52.7) reported by Gargas et al. (1989). However, the blood/air partition coefficient measured for $CHCl_3$ (10.72) was nearly twice that (6.85) reported by Gargas et al. (1989). The experimental results reported in Tables 1 and 2 were conducted at initial liquid concentrations of approximately 100 μg/l. Earlier studies have been conducted at initial liquid concentrations up to 5000 mg/l.

Because of availability, the blood used represented a pooled sample. The measured blood/air partition coefficients were highly reproducible (4–9% as a coefficient of variation). This variability reflects experimental errors in preparation, sampling, and analysis, but not the variability across individuals or across time. In comparison, the variation in human milk, where separate samples from five women were not pooled, was relatively large (14–30%). For example, for $CHCl_3$, donor 4 had a $K_{M/A} = 22.3 \pm 2.5$ ($n = 10$), while donor 5 had $K_{M/A} = 10.5 \pm 0.6$ ($n = 8$) or half the value (Table 3). Some solids and precipitates were observed in samples from these donors, thus, these samples may not have been as well preserved as samples from the other donors. However, milk/air partition coefficients also differed for milk from donors 1, 2 and 3 on different days. No solids or other anomalies were observed in these samples.

4.3. Comparison of cow and breast milk

For cow milk, experimentally determined milk/air partition coefficients were 16.6 ± 1.3 ($n = 7$) for $CHCl_3$, 34.7 ± 1.5 ($n = 8$ for $CHCl_2Br$, 91.2 ± 3.2 ($n = 8$) for $CHClBr_2$ and 257.4 ± 43.9 ($n = 8$) for $CHBr_3$. Measurements of $K_{M/A}$ for human and cow milk were within 3% for $CHCl_3$, $CHCl_2Br$ and $CHClBr_2$. The human milk $K_{M/A}$ for $CHBr_3$ (291.5) was 15% larger than for cow milk (257.4), but the poorer reproducibility of headspace determinations of $K_{M/A}$ for $CHBr_3$ is likely to account for the difference. The lipid content of human and cow milks is very close, and the agreement between experimentally derived partition coefficients suggests that lipid content is key controlling factor.

Human milk can vary in lipid content and possibly other aspects that affect partition coeffi-

Table 3
Human milk/air partition coefficients, $P_{M/A}$, for chloroform using samples from donors 1, 2 and 3 on three different dates and two individual samples from donors 4 and 5

Donor	Date 1	Date 2	Date 3	Average	S.D.
Donor 1	18.00	12.93	13.40	14.78	2.80
Donor 2	13.23	20.39	19.84	17.82	3.98
Donor 3	13.45	13.00	21.45	15.97	4.75
Donor 4	10 replicate determinations on one sample			22.34	2.45
Donor 5	Eight replicate determinations on one sample			10.94	0.57

Table 4
Blood/air and milk/air partition coefficients predicted based on K_{OA} and lipid content

THM	K_{OA} measured/calculated	$K_{B/A}$ based on measured K_{OA}/calculated K_{OA}	$K_{M/A}$ based on measured K_{OA}/calculated K_{OA}
$CHCl_3$	353.8/338.8	2.48/2.37	14.2/13.6
$CHCl_2Br$	954.0/933.3	6.68/6.53	38.2/37.3
$CHClBr_2$	2117.5/2041.7	14.8/14.3	84.7/81.7
$CHClBr_3$	9264.9/5888.4	64.9/41.2	370.6/235.5

'Measured K_{OA}' values were determined experimentally. All experiments based on at least two duplicates. Calculated K_{OA} values are the product of K_{OW} and $K_{W/A}$.

cients. It should be recognized that the milk samples used in the study might not reflect the variability observed in practice. Ideally, simultaneous measurements of milk and blood across a population (random) sample of exposed and lactating women would be used to confirm the laboratory predictions.

4.4. Comparison to predicted values

Table 4 shows the experimentally measured K_{OA} values compared to those derived from K_{OW} and $K_{W/A}$, and the blood/air and milk/air partition coefficients predicted using Eq. (1). Good agreement was found between measured and derived K_{OA} values except for $CHBr_3$, which was underestimated by approximately one-third. For $K_{M/A}$ (milk/air), the experimentally determined results show good agreement with the prediction. However, predictions of $K_{B/A}$ (blood/air) were significantly underestimated compared to experimentally determined values (presented in Table 1), i.e. 2.5/2.4 vs. 10.7 for $CHCl_3$, 6.7/6.5 vs. 26.6 for $CHCl_2Br$, 14.8/14.3 vs. 49.2 for $CHClBr_2$ and 64.9/41.2 vs. 102.3 for $CHBr_3$. This discrepancy may be caused by underestimating the lipid content of blood (0.6% was used) or by the role of blood components other than lipids. No particular lipid fraction is consistent with all the data, however, suggesting that Eq. (1) is fatally flawed. In blood, proteins account for ~7% of plasma, and a portion of these proteins together with blood lipids form a variety of lipoproteins. While strong protein-binding is not expected for THMs, even mild interactions with proteins or other blood components, which are present at high concentrations relative to lipids, could explain results.

Table 5
Partition coefficients predicted using log K_{OW} and lipid content of fluids

THM	Log K_{OW}	$K_{B/W}$	$K_{M/B}$	$K_{B/U}$
$CHCl_3$	1.97	1.6	2.9	1.6
$CHCl_2Br$	2.10	1.9	3.2	1.9
$CHClBr_2$	2.24	2.2	3.6	2.2
$CHBr_3$	2.38	2.7	4.0	2.7

Table 5 shows blood/water, milk/blood, and blood/urine partition coefficients predicted using K_{OW}, the lipid content, and Eq. (2). Errors in $K_{B/A}$ will be propagated when used to estimate liquid–liquid partition coefficients, i.e. milk/blood partition coefficients will be overestimated, and blood/water and blood/urine partition coefficients will be underestimated. Thus, milk/blood partition coefficients were overestimated, and blood/water and blood/urine partition coefficients were underestimated, in comparison to values calculated from experimental data (Table 2). Therefore, $K_{B/A}$ values should not be predicted based solely on the blood lipid content.

4.5. Correlations

Linear relationships between $K_{M/A}$ and K_{OA}, and between $K_{B/A}$ and K_{OA} were observed (Fig. 2), reflecting the hydrophobic nature of the THMs. The following linear regressions were calculated using measured values for the four THMs:

$$\log K_{M/A} = 0.900 \log K_{OA} - 1.095 (R^2 = 0.992) \quad (6)$$

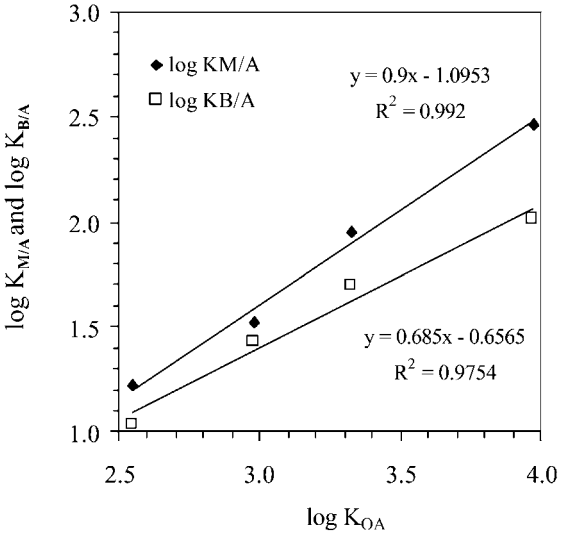

Fig. 2. Relationship between log K_{OA} and log $K_{M/A}$ and log $K_{B/A}$. Regression line and equation indicated on plot.

$$\log K_{B/A} = 0.685 \log K_{OA} - 0.6565 (R^2 = 0.975) \quad (7)$$

These correlations are also supported by observations that levels of other persistent halogenated organic chemicals in milk, blood, adipose tissue and muscles are comparable when calculated on the basis of extractable fat (Jensen, 1991).

Because the THMs are analogous chemicals, linear relationships between log K values are expected with the number of Br atoms (N) in the THM molecule, molecular weight (mol. wt.), and boiling point (bp). For example, the following regressions were estimated using measured values of $K_{B/A}$:

$$\log K_{B/A} = 0.321N + 1.06 (R^2 = 0.994) \quad (8)$$

$$\log K_{B/A} = 0.0072 \text{ mol. wt.} + 0.197 (R^2 = 0.994) \quad (9)$$

$$\log K_{B/A} = 0.0109\text{bp} - 2.584 (R^2 = 0.992) \quad (10)$$

Similar relationships were observed for other measured liquid/air partition coefficients. (These relationships apply only for the THM compounds investigated in this study.) The excellent goodness-of-fit of these regressions, evidenced by high R^2 values, increases confidence in the experimental determinations of the partition coefficients.

5. Discussion

The recommended partition coefficients are listed in Tables 1 and 2. (Predictions in Tables 4 and 5 are primarily for comparison purposes.) These coefficients are suitable for use in PBPK models and other applications.

To varying degrees, all biological samples vary in chemical and physical composition, and measurements of partition coefficients will reflect this variability. Partitioning in human milk, which showed the greatest variability, may be affected by lipid content, protein phases, pH, and other factors (Atkinson and Begg, 1990). Human milk composition varies during the nursing period, i.e. milk contains much less fatty colostrum soon after parturition (Jenson, 1991). Several weeks later, the fat content increases, along with levels of lipophilic contaminants (Mes, 1994). In practice, factors known to modify chemical levels in human milk beyond exposure include the fat content of milk, maternal body weight, duration of lactation, maternal age, and the presence of multiple pregnancies (Sim and McNeil, 1992). These factors, along with the storage concerns noted earlier for a subset of the milk samples, may have increased the variability of the milk measurements.

5.1. Comparison to biological monitoring

A number of studies have performed biological monitoring of THMS, especially for breath and urine. However, few studies have simultaneously measured blood levels, which are necessary to verify experimental determinations. Cammann and Hubner (1995) reported THM levels in blood and urine among competitive swimmers and pool attendants in an indoor pool in Germany. $CHCl_3$ and $CHCl_2Br$ were consistently detected in blood. Concentrations of $CHCl_3$ ranged from 1.2 to 5.2 μg/l in blood and from 9 to 28 μg/l in the pool water. Aggazzotti et al. (1995) summarized find-

ings among swimmers at 12 indoor pools in Italy, finding $CHCl_3$ levels from 23 to 114 (mean of 31) $\mu g/l$ in water, 48–459 (mean of 214) $\mu g/m^3$ in air, 14–312 (mean of 94) $\mu g/m^3$ in alveolar air, and 0.1–3 (mean of 1.1) $\mu g/l$ in plasma. In a later study, Aggazzotti et al. (1998) showed that 1 h of swimming in an indoor pool with a 34 ± 10-$\mu g/l$ of chloroform resulted in alveolar air concentrations of 77 ± 19 $\mu g/m^3$, and plasma concentrations of 1.5 ± 0.5 $\mu g/l$. (Based on the K_{OW} and lipid content, levels in whole blood are expected to be 50–100% higher than in plasma.)

Partition coefficients for THMs cannot be established using the studies cited above for several reasons. First, experimental results may not represent equilibrium due to relatively rapid clearance of the THMs. The timing of samples relative to exposure is a critical factor in understanding the relationship of biological monitoring results to short-term exposures (Batterman et al., 1998). Nonetheless, the mean concentrations reported in the two studies by Aggazzotti et al. cited above give plasma/alveolar air ratios of 11.7 and 19.5, quite close to the $K_{B/A}$ value of 10.7 determined here. Second, the overall dose is not precisely known since swimmers experience THM uptake via multiple pathways (inhalation, dermal absorption, and incidental ingestion), and the 'administered' dose rate varied. Third, measurements are limited, e.g. studies have focused on $CHCl_3$, other THMs have received less attention, and human milk has not been studied quantitatively.

5.1.1. THM levels in human milk

Like other chemicals, the distribution of the THMs among biological media is determined by water and lipid solubility, ease of metabolism, storage in the body, excretion, and transfer of the chemical into various compartments (Wolf, 1983; Jensen, 1991; Sim and McNeil, 1992). Since THMs are cleared relatively rapidly, an additional factor in determining lactational transfer is the timing of exposure and feeding events. Lactational transfers are potentially important since over half (52%) of mothers in the US breast-fed their infants in 1989, 40% continued for 3 months or more (NAS, 1991), and overall 22% of infants under 1 year are breast-fed (Maxwell and Burmaster, 1993).

In this study, the milk/blood partition coefficients for the more brominated THMs approached 3, suggesting that lactational transfer might be important. However, THMs are both stored and metabolized during milk production and the interval to milk consumption, and relatively fast clearance will lower THM levels in blood following exposure. Either direct measurement in exposed and lactating women or analysis using physiologically-based pharmacokinetic (PBPK) lactation models is needed to account for metabolism, storage, timing of exposure and feeding events, and other factors that govern exposure to the nursing infant. While such models have been used to estimate the ingestion rates of some chemicals by nursing infants (Shelley et al., 1988; Hoover et al., 1991; Schrieber, 1993; Byckowski et al., 1993; Fisher et al., 1997), no such study has been performed for the THMs. The partition coefficients reported here can be used in PBPK models. While magnification of THMs in human milk compared to levels in blood is suggested by the equilibrium analysis, the assumption of equilibrium conditions will overestimate contaminant levels (Filser and Bolt, 1981) and a PBPK model analysis is needed. Ideally, simultaneous blood and milk measurements would be collected and analyzed for PBPK model confirmation.

6. Conclusions

Experimentally determined partition coefficients for the THMs for blood/air range from 10.7 to 102.3, for milk/blood from 1.3 to 2.9, and for blood/urine from 3.4 to 4.9, depending on the compound. These results are relevant to concentrations attributable to environmental exposures. Observed and predicted partition coefficients for milk/air were similar, but partition coefficients involving blood differed. The expected correlations involving partition coefficients and the structure and characteristics of THMs were noted, increasing confidence in the experimental results.

Results for human and cow milk were similar. The experimentally-determined partition coefficients can be used to help interpret results of biological monitoring aimed at quantifying THM exposure, and they are suitable for use in PBPK models for estimating maternal and infant exposures.

Acknowledgements

The authors thank the US EPA for partial financial support of this research.

References

Aggazzotti G, Fantuzzi G, Righi E, Predieri G. Environmental and biological monitoring of chloroform in indoor swimming pools. J Chromatogr A 1995;710(1):181–190.

Aggazzotti G, Fantuzzi G, Righi E, Predieri G. Blood and breath analyses as biological indicators of exposure to trihalomethanes in indoor swimming pools. Sci Total Environ 1998;217:155–163.

Albritton EC, editor. Standard values in blood. Philadelphia and London: W.B. Saunders Co, 1952.

Arora H, LeChevallier MW, Dixon KL. DBP occurrence survey. J Am Water Works Assoc 1997;89:60–68.

Atkinson HC, Begg EJ. Prediction of drug distribution into human milk from physiochemical characteristics. Clin Pharmacokinetics 1990;18(2):151–167.

Ayotte P, Larue M, Riopel A, Messier F. Formation des trihalomethanes dans les systems de faible capacite. In: Toft P, Tobin RS, Sharp Toft J (Eds). Drinking water treatment: small system alternatives. Proceedings of the Third National Conference on Drinking Water, St. John's, Newfoundland, Canada, June 12 to 14, 1988.

Batterman S, Franzblau A, D'Arcy JB, Sargent NE, Gross KB, Schreck RM. Breath, urine and blood measurements as biological exposure indices of short-term inhalation exposures to methanol. Int Arch Occup Environ Health 1998;71:325–335.

Byckowski JZ, Kinkead ER, Greene RJ, Bankston LA, Fisher JW. Physiological based modeling of the lactational transfer of tetrachloroethylene. Toxicologist 1993;13:354.

Byckowski JZ, Gearhardt JM, Fisher JW. Occupational exposure of infants to toxic chemicals via breast milk. Nutrition 1994;10(1):43–48.

Cammann K, Hubner K. Trihalomethane concentrations in swimmers' and bath attendants' blood and urine after swimming or working in indoor swimming pools. Arch Environ Health 1995;50:61–65.

Craan AG, Haines DA. Twenty-five years of surveillance for contaminants in human breast milk. Arch Environ Contamin Toxicol 1998;35(4):702–710.

Craun GF. Drinking water disinfection: assessing health risks. Health Environ Digest 1990;4:1.

Cumming RB, Jolley RL. Occurrence and exposures to disinfection by-products. In: Craun GF, editor. Safety of water disinfection: balancing chemical & microbial risks. Washington DC: ILSI Press, 1993:257–275.

Dewailly E, Ayotte P, Lialiberte C, Weber J-P, Gingras S, Nantel AJ. Polychlorinated biphenyl (PCB) and dichlorodiphenyl dichloroethylene (PDDE) concentrations in the breast milk of women in Quebec. Am J Pub Health 1996;86:1241–1246.

Filser JG, Bolt HM. Inhalation pharmacokinetics based on gas uptake studies. I. Improvement of kinetic models. Arch Toxicol 1981;47:279–292.

Fisher J, Mahle D, Bankston L, Greene R, Gearhart J. Lactational transfer of volatile chemicals in breast milk. Am Ind Hygiene Assoc J 1997;58:425–431.

Gargas ML, Burgess RJ, Voisard DE, Carson GH, Andersen ME. Partition coefficients of low- molecular-weight volatile chemicals in various liquids and tissues. Toxicol Appl Pharmacol 1989;98:87–99.

Gladen BC, Rogan WJ, Hardy P, Thullen J et al. Development after exposure to polychlorinated biphenyls and dichlorodiphenyl dichloroethylene transplacentally and through human milk. J Pediat 1988;113:991.

Guyton AC. Textbook of medical physiology. 8th edition Philadelphia: W.B. Saunders Company, Harcourt Brace Jovanovich, Inc, 1991:915–928.

Hansch C, Leo AJ. Medchem Project Issue 26. Claremont, CA: Pomona College, 1985.

Harner T, Bidleman TF. Measurements of octanol–air partition coefficients for polychlorinated biphenyls. J Chem Eng Data 1996;41:895–899.

Hoover S, Zeise L, Krowech G. Exposure to contaminants through breast milk. In: Garrick BJ, Gekler WC, editors. The analysis, communication, and perception of risk. London: Plenum Press, 1991:257.

Jacobson JL, Jacobson SW, Humphrey HEB. Effects of exposure to PCBs and related compounds on growth and activity in children. Neurotoxicol Teratol 1990;12:319–326.

Jensen AA. Transfer of chemical contaminants into human milk. In: Chemical Contaminants in Human Milk. Jensen AA, Slorach SA, Eds. Chemical contaminants in human milk. CRC Press, Boca Raton, FL, 1991.

Jo W, Weisel C, Lioy P. Chloroform exposure and the health risk associated with multiple uses of chlorinated tap water. Risk Anal 1990;10(4):581–585.

Jorgenson TA, Meierhenry EF, Rushbrook CJ, Bull RJ, Robinson M. Carcinogenicity of chloroform in drinking water to male Osborne–Mendel rats and female B6C3F1 mice. Fund Appl Toxicol 1985;5:760–769.

Koppe JG, Pluim E, Olie K. Breast milk, PCBs, dioxins, and vitamin K deficiency: discussion paper. J R Soc Med 1989;82:416.

Mabe WR, Smith JH, Podoll RT et al. Aquatic fate process data for organic priority pollutants, EPA 440/4-81-013, PB87-169090. Washington DC: US Environmental Protec-

tion Agency, Office of Water Regulations and Standards, 1982.
Maxwell NI, Burmaster DE, Ozonoff D. Trihalomethanes and maximum contaminant levels: The significance of inhalation and dermal exposure to chloroform in household water. Reg Toxicol Pharmacol 1991;14(3):297–312.
Maxwell NI, Burmaster DE. A simulation model to estimate a distribution of lipid intake from breast milk during the first year of life. J Exposure Assess Environ Epidem 1993;3: 383–406.
McGuire MJ. Preparing for the disinfection by-products rule: a water industry status report. J Am Water Works Assoc 1989;81:35–40.
McGuire MJ, Meadow RG. Trihalomethane survey. J Am Water Works Assoc 1988;80(1):61.
McKone TE. Linking a PBPK model for chloroform with measured breath concentrations in showers: implications for dermal exposure models. J Exposure Assess Environ Epid 1993;3(3):339–365.
Mes J. Temporal changes in some chlorinated hydrocarbon residue levels of Canadian breast milk and infant exposure. Environ Pollution 1994;84(3):261–268.
NAS. Nutrition during lactation. Washington, DC: National Academy of Sciences, National Academy Press, 1991.
NCI. Report on the carcinogenesis bioassay of chloroform [NTISPB-PB-264-018]. Bethesda, MD: National Cancer Institute, 1976.
Newsome WH, Davies D, Doucet J. PCB and organochlorine pesticides in Canadian human milk. Chemosphere 1995;30(11):2143–2153.
NTP (1985) Toxicology and carcinogenesis studies of chlorodibromomethane (CAS No. 124–48- 1) in F344/N rats and B6C3F mice (gavage studies). National Toxicology Program Tech Report Series No. 282. NTIS PB 86-166675.
NTP (1986) Studies of bromodichloromethane in F344/N rats and B6C3F1 mice (gavage studies), Technical report no. 321, NIH Publ. No. 87-2537, National Toxicology Program.
NTP (1987) NTP Technical Report on the Toxicology and Carcinogenesis Studies of Bromodichloromethane (CAS No. 75-27-4) in F344/N Rats and B6C3F1 Mice (Gavage Studies). National Toxicology Program Tech. Report Series no. 321. US Dept. Health and Human Services, Public Health Service, National Institute of Health.
NTP (1988) Technical Report on the Toxicology and Carcinogenicity Studies of Tribromomethane(Bromoform) in F344 rats and B6C3F1 mice (gavage study). National Toxicology Program TR-350.
Olin SS, editor. Exposure to contaminants in drinking water. New York: CRC Press, 1998:232.
Pellizzari ED, Hartwell TD, Harris III BSH, Waddell RD, Whitaker DA, Erickson MD. Purgeable organic compounds in mother's milk. Bull Environ Contam Tox 1982; 28:322–328.
Ryan AS, Rush D, Krieger FW, Lewandowski GE. Recent declines in breast feeding in the United States, 1984–1989. Pediatrics 1991;88:719–727.
Sato A, Nakajima T. Partition coefficients of some aromatic hydrocarbons and ketones in water, blood and oil. Br J Ind Med 1979;36:231–234.
Schrieber JS. Predicted infant exposure to tetrachloroethylene in human breast milk. Risk Anal 1993;13(5):515.
Shelley ML, Andersen ME, Fisher JW. An inhalation distribution model for the lactating mother and nursing child. Toxicol Lett 1988;43:23–29.
Sim MR, McNeil JJ. Monitoring chemical exposure using breast milk: a methodological review. Am J Epi 1992; 11136(1):1–11.
US EPA (1997a) Integrated Risk Information System, IRIS substance files for chloroform (4/1/97), bromodichloromethane (3/1/97), dibromochloromethane (3/1/97), bromoform (3/1/97 , accessed online.
US EPA (1997b) Exposure Factors Handbook, US Environmental Protection Agency, Office of Research and Development, Washington, DC. Aug.
Vander AJ, Sherman JH, Luciano DS, editors. Human physiology: the mechanism of body function, 5th edition. New York: McGraw-Hill Pub. Co, 1990.
Weisel C, Jo W. Ingestion, inhalation, and dermal exposure to chloroform and trichloroethene from tap water. Environ Health Persp 1996;104(1):48–51.
Wolf MS. Occupationally derived chemicals in breast milk. Am J Ind Med 1983;4:259–281.